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ANSWER 1 OF 2 BIOSIS COPYRIGHT 2001 BIOSIS

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TI Method and kits for preparing multicomponent nucleic acid constructs.

ΑU Harney, Peter D. (1)

(1) Aliso Viejo, CA USA CS

ASSIGNEE: VectorObjects, LLC, Wellesley, MA, USA

PΙ US 6277632 August 21, 2001

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DTPatent

LΑ English

The invention provides a highly efficient, rapid, and cost effective AB method of linking nucleic acid components in a predetermined order to produce a nucleic acid multicomponent construct. The invention further provides nucleic acid components, each nucleic acid component comprising a double stranded nucleic acid molecule having at least one single stranded 5' or 3' terminal sequence, the terminal sequence having sufficient complementarity to either a terminal sequence in a separate nucleic acid component or to a sequence in a linking nucleic acid molecule so as to allow for specific annealing of complementary sequences and linkage of the

components in a predetermined order. Kits containing reagents required to practice the method of the invention are also provided.

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L2ANSWER 2 OF 2 CAPLUS COPYRIGHT 2001 ACS

AN 1998:31322 CAPLUS

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ligation of nucleic acid components with complementary terminal sequences IN Harney, Peter D. PA Biodynamics Associates, USA SO PCT Int. Appl., 33 pp. CODEN: PIXXD2 DT Patent LA English FAN.CNT 1 KIND DATE APPLICATION NO. DATE PATENT NO. _ _ _ _ _____ ______ ______ ΡI WO 9748716 **A**1 19971224 WO 1997-US10523 19970616 W: AL, AM, AT, AU, AZ, BA, BB, BG, BR, BY, CA, CH, CN, CZ, DE, DK, EE, ES, FI, GB, GE, GH, HU, IL, IS, JP, KE, KG, KP, KR, KZ, LC, LK, LR, LS, LT, LU, LV, MD, MG, MK, MN, MW, MX, NO, NZ, PL, PT, RO, RU, SD, SE, SG, SI, SK, TJ, TM, TR, TT, UA, UG, UZ, VN, YU, ZW, AM, AZ, BY, KG, KZ, MD, RU, TJ, TM RW: GH, KE, LS, MW, SD, SZ, UG, ZW, AT, BE, CH, DE, DK, ES, FI, FR, GB, GR, IE, IT, LU, MC, NL, PT, SE, BF, BJ, CF, CG, CI, CM, GA, GN, ML, MR, NE, SN, TD, TG CA 2258570 AA19971224 CA 1997-2258570 19970616 AU 9733997 **A**1 19980107 AU 1997-33997 19970616 EP 915903 **A1** 19990519 EP 1997-930083 19970616 AT, BE, CH, DE, DK, ES, FR, GB, GR, IT, LI, LU, NL, SE, MC, PT, IE, FI JP 2000512852 T2 20001003 JP 1998-503272 19970616 US 6277632 В1 20010821 US 1997-877034 19970616 PRAI US 1996-19869 Р 19960617 WO 1997-US10523 W 19970616 The invention provides a highly efficient, rapid, and cost effective AB method of linking nucleic acid components in a predetd. order to produce a nucleic acid multicomponent construct. The invention further provides nucleic acid components, each nucleic acid component comprising a double-stranded nucleic acid mol. having at least one single-stranded 5' or 3' terminal sequence, the terminal sequence having sufficient complementarity to either a terminal sequence in a sep. nucleic acid component or a sequence in a linking nucleic acid mol. so as to allow for specific annealing of complementary sequences and linkage of the components in a predetd. order. The various nucleic acid components can be linked via, without limitation, the following: (1) annealing of 5' complementary terminal sequences in 2 sep. nucleic acid components; (2) annealing of 3' complementary terminal sequences in 2 sep. nucleic acid components; (3) annealing of an oligonucleotide bridge with complementary 5' and 3' terminal sequences in 2 sep. nucleic acid components; or (4) annealing of an adaptor mol. with complementary 5' or 3' terminal sequences in 2 sep. nucleic acid components;. To demonstrate the simultaneous assembly of multiple nucleic acid components having unique, non-palindromic terminal sequences, to produce a viable plasmid vector, 3 nucleic acid components were used: a gene coding for green fluorescent protein; a 0.6-kb mol. coding for terminator sequences and a histidine tag; and a 2.5-kb mol. coding for the lac promoter, an ampicillin resistance gene, and an origin of replication. This method is

particularly suitable for the construction of nucleic acid vectors.

Preparation of multicomponent nucleic acid constructs by

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